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strict compliance with the recommendations of the commissioners on uniform laws, to the effect that uniformity in food and drug laws can best be secured by amendments to the State laws with a view of bringing them in complete harmony with the requirements of the Federal law.

Some of the more recently enacted laws, however, appear to demonstrate the impracticability of this suggestion and the need for additional safeguards in the way of standards for foods and food accessories.

To provide uniform standards, an effort is being made at the present time under the auspices of several organizations of men interested in the enforcement of food and drug laws, to outline and to offer for general adoption suitable standards for food products.

The food and drug laws of a number of States and Territories have been amended during the past year. Prominent among the laws that have been so modified are those of Hawaii, Iowa, Massachusetts, Nebraska, North Carolina, North Dakota, Oklahoma, Rhode Island, Vermont, and Wyoming.

In Hawaii the definition for the term "food" is made "to include all articles used for food, drink, confectionery, or condiment by man or animals, whether simple, mixed, or compound."

In Iowa "food" is defined as including all articles used for or entering into the composition of food, drink, confectionery, or condiment used by man or domestic animals.

An amendment to the pure-food law of Nebraska defines commercial feeding stuffs and makes provisions for their analysis.

The amended pure-food law of North Dakota outlines standards for a number of articles and includes a number of sections in which the requirements are presented in a negative form.

VITAMINES AND NUTRITIONAL DISEASES.

A STABLE FORM OF VITAMINE, EFFICIENT IN THE PREVENTION AND CURE OF CERTAIN NUTRITIONAL DEFICIENCY DISEASES.¹

By ATHERTON SEIDELL, Technical Assistant, Hygienic Laboratory, United States Public Health Service.

According to the recently developed conceptions of nutritional deficiency diseases, particularly beriberi and pellagra, a factor, hitherto unrecognized, must be taken into consideration in respect to the essential food elements necessary for normal metabolism. This constituent has been given the name "vitamine," and this term now expresses the idea that in addition to the usual proteins, fats, carbohydrates, etc., contained in ordinary foodstuffs there must be

present also a sufficient amount of vitamine in order that normal metabolism be maintained.

It is recognized that although vitamines undoubtedly are widely distributed in food products, they occur for the most part in very minute amounts, and the various foods differ in the proportions which they contain. In the case of an ordinary mixed diet a supply of vitamines ample for the needs of the organism will no doubt usually be present. On the other hand, if the diet is made up principally of foods poor in vitamines or rendered so by their preparation, it would be expected that an insufficient amount of these substances would be provided and that abnormal metabolic processes would result. The studies which have been made on beriberi, and especially on polyneuritis in fowls, have definitely established this conception of the rôle of vitamines in metabolism.

In all attempts which have so far been made to isolate vitamines in sufficient quantities for experimental studies, only very small yields have been obtained. This has been due to the apparent destruction of the physiologically active substance during the various steps of the several processes employed. On this account it has so far not been possible to make convincing demonstrations of the specific function of isolated vitamines in nutritional deficiency diseases of human beings. It was in the hope of developing an efficient procedure for obtaining a relatively concentrated and comparatively cheap form of vitamine, suitable for studies on the prevention and cure of such human nutritional deficiency diseases as beriberi, pellagra, infantile malnutrition, etc., that the following briefly-described experiments were undertaken:

Of the various substances which have been shown to be rich in vitamines, brewer's yeast is probably, at present, the cheapest. In all save perhaps a few of the larger brewing establishments the bottom yeast remaining after each brew is washed down the sewer. This material, therefore, appeared to meet every requirement for cheapness. As obtained from the brewery it is in the form of a light-colored mush. It is first subjected to pressure in a hydraulic press for removal of the beer still retained by it. The press cake is then stirred with ice water and again pressed out. The cake is placed in enamel-ware vessels and brought to a temperature of 37.5° C. and held there for a period of about 48 hours. The autolysis is more or less complete at the end of this time and the material converted to the consistency of thick soup. After cooling to room temperature, the liquid is filtered through large folded filter papers. All methods of filtration involving the use of pressure which were

¹Acknowledgments are due the Christian Heurich Brewing Co., of Washington, for the very liberal supplies of yeast used in the present investigation.

tried, failed. The weight of clear red brown filtrate amounts to approximately 50 per cent of the weight of the unfiltered material.

This filtered autolyzed yeast contains about 23 per cent of total solids, determined by evaporation in a vacuum desiccator. It will keep in a cool place, without noticeable change, for several months, possibly much longer. If administered in 1 c. c. doses on alternate days to a pigeon kept on an exclusive diet of polished rice, the pigeon does not lose weight nor show symptoms within at least two months, the period during which the present experiment has been under way. A pigeon kept on polished rice without the yeast filtrate begins to lose weight usually within the first 5 days and dies with the typical paralysis of polyneuritis within about 20 days. If 1 c. c. of the yeast filtrate is given to completely paralyzed pigeons a relief of the paralysis will occur within an hour and to all outward appearances the pigeon will be restored to a normal condition within 12 hours.

Although this yeast filtrate is evidently very active and has good keeping qualities, an effective dose for a man, as estimated from the amount required for pigeons, would be about 200 c. c. It was therefore realized that the material would have to be considerably concentrated before it could be successfully used in human cases.

The procedures originally employed for concentrating the veast filtrate included vacuum distillation, evaporation in a current of dry air, and freezing and removing the ice by centrifugation according to the method perfected by Mr. H. C. Gore, of the Bureau of Chemistry, for concentrating cider and fruit juices. Although a portion of the dissolved solids separated during the concentration and could be removed, the resulting concentrated liquid became more and more sirupy and it was impossible to convert the material to a solid condi-The use of alcohol for precipitating the inactive material from the very concentrated solution was at first thought to be a practical step, since an easily removed granular product was obtained. concentrating the alcoholic filtrate from this product and estimating its activity by experiments on neuritic pigeons it was found that a considerable loss of the active constituent had occurred. An examination of the solid precipitated by the alcohol showed it to be quite active; hence a distribution of the vitamine had taken place. It was therefore apparent that the use of alcohol for concentrating the vitamine in yeast filtrate was not a practical procedure from an economic standpoint.

After experiments with a number of other procedures had failed to yield satisfactory results, attention was turned to the possibility of using a medium which might exert a selective adsorption for the vitamine. It was shown several years ago by Prof. John Uri Lloyd, of Cincinnati, that certain varieties of fuller's earths could, by proper treatment, be rendered very efficient in the selective adsorption of

alkaloids from complex mixtures. It was thought that possibly this material might remove the vitamine from the autolyzed yeast filtrate. Preliminary experiments indicated that such a removal of it could be effected. After beginning the work, there was found a published statement by Chamberlain and Vedder (Philipp, J. Sci. (B) 6, 399, 1911) to the effect that the neuritis-preventing substance of extracts of rice polishings is removed by filtering through bone black. This observation strengthened the expectation that a separation of the vitamine from the substances accompanying it in filtered autolyzed yeast could be effected by means of an efficient adsorptive agent.

Through the kindness of Prof. Lloyd a large quantity of the especially selected and prepared hydrous aluminum silicate which he uses for alkaloidal separations and which is known as "Lloyd's Reagent," was sent to the writer. It was decided that in the first experiment a relatively large amount of the adsorptive agent should be used. Two hundred grams per liter of yeast filtrate were therefore selected, and the mixture was thoroughly shaken during about 15 minutes and then filtered through a large Buchner funnel. The filtrate was reserved and the residue pressed out with a spatula and, without washing, drained with air suction as thoroughly as possible. The solid residue was transferred to a vacuum desiccator containing concentrated H_2SO_4 and dried to constant weight.

As indicated in connection with the description of the autolyzed yeast filtrate, there are two methods of determining the activity of a product in respect to polyneuritis—first by the prevention of the onset of the symptoms and second by means of curative experiments. Both methods were employed in the case of the solid, which had been shaken with the autolyzed yeast filtrate, and of the clear liquid recovered after treatment with the solid.

In the case of the curative experiments it was found that a pigeon suffering from severe paralysis due to an exclusive diet of polished rice, showed unmistakable improvement within an hour after a dose of the solid corresponding to 3 c. c. of the yeast filtrate, and to all outward appearances was normal the next morning. Similar experiments made with the filtrate from the solid showed it to have no appreciable curative action.

The feeding experiments were conducted on a group of 12 pigeons. They were kept in a large cage and given as much polished rice as they would eat. They were weighed and dosed as follows at intervals of two to three days: Two of the pigeons were given 3 c. c. doses of the original yeast filtrate (Y. F.), three received doses of 0.6 gram of the activated solid (A. S.) corresponding to 3 c. c. of Y. F., three received 3 c. c. doses of the filtrate from the activated solid (F. A. S.), and four were retained as controls.

The results are plotted on the accompanying chart. It will be seen that the pigeons receiving the autolyzed yeast filtrate and the solid activated by being shaken with the yeast filtrate, not only showed no ill effects from the exclusive diet of polished rice but actually gained considerably in weight. The controls and those receiving the filtrate from the activated solid all developed the typical paralysis of polyneuritis within from 11 to 23 days. The experiment, therefore, proves conclusively that, using a ratio of 200 grams per liter, the adsorptive agent removes the vitamine promptly and completely from autolyzed yeast filtrate.

Attention was next turned to ascertaining how small a proportion of the solid reagent would adsorb the vitamine completely from the

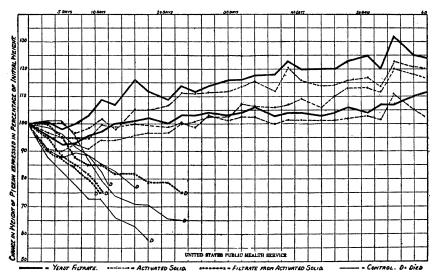


Chart showing effect of yeast filtrate, activated solid, and filtrate from activated solid on pigeons kept upon an exclusive diet of polished rice. (Dosage began on fifth day.)

yeast liquor. This information was considered desirable in order that an activated solid containing as high a concentration of the vitamine as possible might be obtained; one in which doses sufficient for human patients would be contained in amounts of the solid small enough for convenient administration.

An experiment made with the solid and with the filtrate obtained by separating a mixture of only 20 grams of the reagent to 1 liter of yeast hquor showed that, although the solid was active when given in doses corresponding to 3 c. c. of the original yeast liquor, the liquid filtered from the solid still retained a sufficient amount of vitamine to protect pigeons for periods of over two months when given in 3 c. c. doses on alternate days. From this it is evident that only an incomplete separation had been effected.

The proportion of 40 grams of solid per liter was next used but the filtrate was found still to be just slightly active. When 50 grams per liter were used only an inconsiderable amount of vitamine appeared to remain in the yeast filtrate. This, therefore, was considered to be the lowest proportion of solid reagent which could be employed for removing the active material from autolyzed yeast filtrate. A curative experiment made with the filtrate obtained in using 50 grams solid per liter resulted as follows: A pigeon which was beginning to show weakness indicative of the approach of the paralysis of polyneuritis was given 5 c. c. of the filtrate and the next day showed a very slight improvement. The 5 c. c. doses of the filtrate were continued on alternate days, but the symptoms of polyneuritis developed progressively until typical paralysis, followed by death, resulted on the eighth day succeeding that of the first dose. There was evidently just enough active material in the relatively larger doses of filtrate to delay the fatal termination about one week.

On the basis of the above-described experiments the procedure which has been adopted for preparing activated solid from autolyzed yeast liquor is as follows: To a large volume of clear autolyzed yeast filtrate is added 50 grams per liter of the colloidal hydrous aluminium silicate reagent as prepared by Prof. Lloyd, of Cincinnati, for alkaloidal separations. The mixture is well shaken and allowed to stand several hours until the supernatant liquid is practically free of suspended solid. The dark liquid is then siphoned off and about an equal volume of water and enough standard hydrochloric acid is added to yield an approximately N/100 acid solution. The addition of this small amount of acid is necessary to hasten subsidence of the solid. After shaking and allowing to stand, the supernatant liquid is again siphoned off. The solid is washed a second time with the very dilute acid and then poured upon a large Buchner funnel and washed several times with small portions of water and finally with about three small portions of 95 per cent alcohol. The solid is then spread out on paper to permit the evaporation of the major part of the alcohol and finally dried to constant weight in vacuum desiccators containing concentrated sulphuric acid.

Both preventive and curative experiments on pigeons have been made with this material, and the results agree with those already described and illustrated by the chart for the product made with the ratio of 200 grams of reagent per liter of yeast filtrate. It has been found that prompt and effective cures of completely paralyzed pigeons result from 0.05 gram doses of the activated solid, corresponding to 1 c. c. of the original yeast filtrate. Preventive experiments, continued at present for over a month, using 0.05 gram of the solid on alternate days, show that the pigeon retains its normal health and weight on an exclusive diet of polished rice.

On the basis of 60 kilograms as the weight of a man, as compared with 300 grams for the pigeon, a comparable dose of the activated material would be 10 grams of the solid on alternate days, or 5 grams per day. This is a quantity which could be conveniently taken either in capsules or as an aqueous suspension. The material is practically tasteless and odorless, and aside from the vitamine which it contains is an absolutely inert substance which would produce no noticeable effects on passage through the body.

In regard to the probable therapeutic value of the product here described, it should be mentioned that much evidence has been advanced to show that polyneuritis in birds is essentially the same disease as beriberi in man, consequently, it may be expected that the concentrated vitamine may prove to be an efficient remedy for beriberi. That it will be valuable in the treatment of pellagra can not be asserted until clinical tests on human cases have been made; it can only be said that if the source of the vitamine at present selected—viz, yeast—should happen not to be well chosen as regards pellagra, the method here described for concentrating the yeast vitamine probably could be applied equally effectively to some other raw product.

In conclusion it should be mentioned that the procedure here developed for yeast can be used with slight modifications in the estimation of the vitamine content of various food products. Experiments already under way upon the potato have demonstrated the applicability of the method.

PLAGUE-PREVENTION WORK

CALIFORNIA.

The following report of plague-prevention work in California for the week ended January 22, 1916, was received from Surg. Boggess, of the United States Public Health Service, in charge of the work:

SAN FRANCISCO, CAL.	1	SAN FRANCISCO, CAL.—Continued.	
RAT PROOFING. New buildings:		RAT PROOFING—continued.	
Inspections of work under construction.	187	Old buildings-Continued.	
Basements concreted (square feet		Yards and passageways, planking re-	
59,680)	70	moved	35
Floors concreted (square feet, 19,670)	13	Cubic feet new foundation walls in-	
Yards, passageways, etc. (square feet,		stalled	8,800
25,664)	109	Concrete floors installed (square feet,	
Total area of concrete laid (square feet).	105,014	30,162)	44
Class A, B, and C (fireproof) buildings:	100	Basements concreted (square feet,	•
Inspections made	129	36,725)	43
Roof and basement ventilators, etc., screened	547	Yards and passageways, etc., concreted	
Wire screening used (square feet)	2,650	(square feet, 15,060)	92
Openings around pipes, etc., closed	2,000	Total area concrete laid (square feet)	81,947
with cement	911	Floors rat proofed with wire cloth	
Sidewalk lens lights replaced	800	(square feet, 350)	. 1
Old buildings:		Buildings razed	19
Inspections made	503	New garbage cans stamped approved	598
Wooden floors removed	43	Nuisances abated	297